

Surface Induced Dissociation in a Quadrupole-Time of Flight Mass-Spectrometer

Tandem mass spectrometry is a well established analytical technique for the characterization and identification of molecular structures, and explores action of gas phase ion chemistry. This technique involves activation of selected precursor ions followed by mass analysis of the fragment ion products. The nature of ion fragmentation depends on the method of ion activation. The most common activation method, collision-induced dissociation (CID) or collisionally-activated dissociation (CAD), involves precursor ion excitation via multiple collisions with an inert gas. The efficiency of CID is dependent on the ratio between the relative masses of the target gas and the colliding ion. The lower the mass of the target gas, the lower the collision energy in the center of mass reference frame. Traditionally most CID targets have been low mass inert gases such as He or Ar, therefore even with multiple collisions, the total energy conversion is limited. This property sometimes limits the utility of CID when dealing with large, complex biological molecules such as intact proteins or even non-covalent complexes, which require high activation energy to induce dissociation.

Surface induced dissociation (SID) is an alternative collisional ion activation method, first introduced for organic compounds by Cooks and co-workers. This technique has been used by several research groups to analyze small molecules, peptides and even proteins. Ion activation in SID is analogous to CID; except that a target surface often an organic thin film self assembled monolayer (SAM) is used as a collision target instead of inert gas. SID more closely approximates a single collision process compared to multiple collisions in CID and believe to deposit more internal energy upon collision.

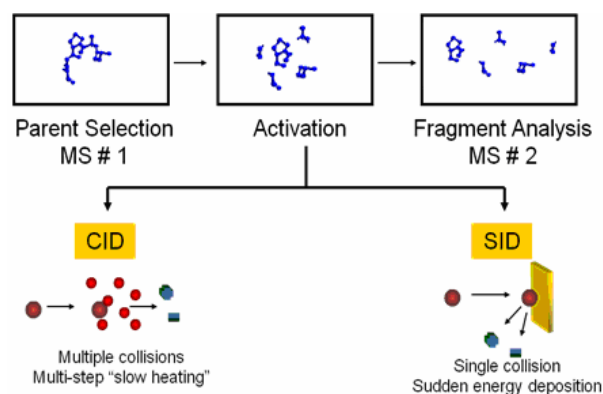


Figure 1: CID (multiple collision vs. SID (single collision)

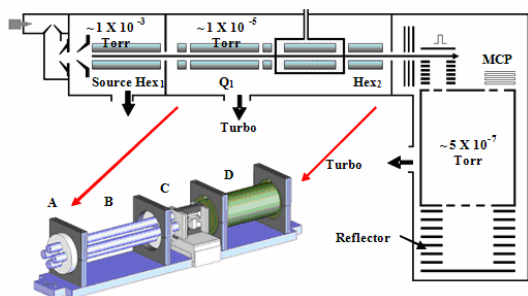


Figure 2: Modified Q-TOF instrument to accommodate inline SID device

Although significant progress has been made in the development of SID instrumentation and the understanding of SID collision processes, the application of SID as a routine and effective activation method in commercial mass spectrometers has yet to be realized. Our goal was to incorporate an SID device into a quadrupole time of flight mass spectrometer with minimal instrument modification so as not to disturb the original instrument performance into the original configuration of this commercial instrument (Figure 2).

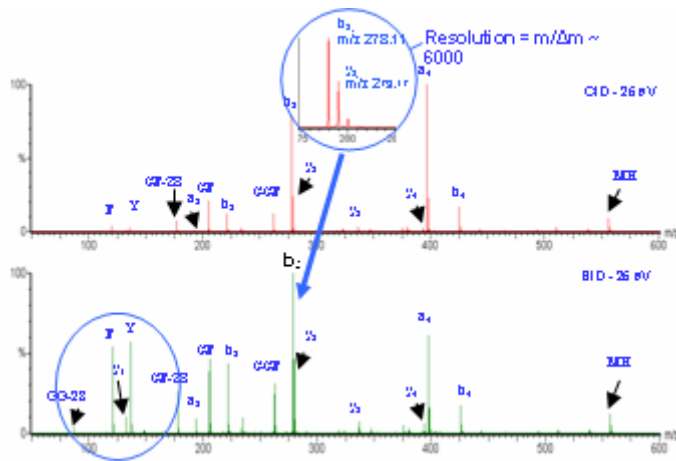


Figure 3: YGGFL, CID vs. SID comparison

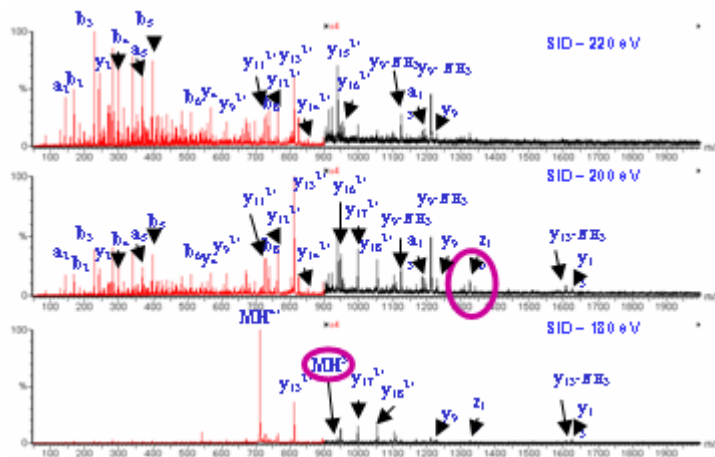


Figure 4: Melittin (GIGAVLKVLTTGLPALISWIKRKR QQ-NH₂+4H)⁴⁺ at different SID energies

The best performing in-line SID design consists of three main functional regions; ion focusing region, surface region and fragment ion extraction region. The ion beam coming out of the quadrupole is first focused by three entrance lenses and deflected upward, off-axis, by a bottom “U” deflector and a top flat/slanted deflector onto the surface to undergo SID.

A broad range of samples, from low mass peptides up to a range of mass ~ 215 kDa, including YGGFL, RPPGFSPF, angiotensin I, II, III, fibrinopeptide A and B, insulin B chain, intact Insulin, melittin, glucagons, ubiquitin, cytochrome C and some large non-covalent complexes such as cytochrome C dimers and heat shock proteins were successfully fragmented with the new SID setup. In comparison with CID of peptides, some differences were noticed with SID spectra showing greater b ion abundance, higher abundance of immonium ions, charge stripping behavior of multiply charge parent ions and energy shifts in some fragmentation pathways.